EFFECTS OF AERIAL APPLICATION OF ZECTRAN® UCIF 19 ON THE GROWTH AND DEVELOPMENT OF NESTLING WHITE-THROATED SPARROWS

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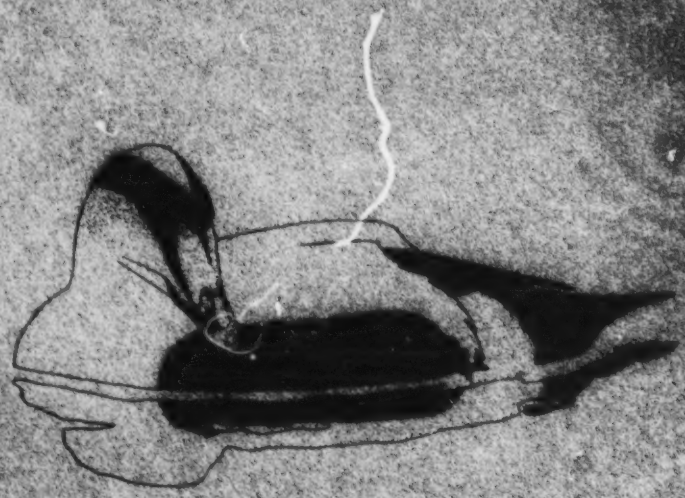

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Abstract

The cholinesterase-inhibiting insecticide Zectran® is currently being tested for spruce budworm (Choristoneura fumiferana) control in Canada. Because of its relatively high acute oral toxicity to birds, concern about Zectran's registration prompted the Canadian Wildlife Service (Atlantic Region) to determine if field application of it is hazardous to songbirds. Of particular concern are nestlings, which are more sensitive to cholinesterase-inhibiting insecticides than are adult songbirds.

Growth and development of White-throated Sparrow nestlings were studied in an area subjected to aerial application of Zectran® (a carbamate insecticide, active ingredient mexacarbate). Comparison of daily means between exposed and control nestlings of four parameters, weight, tarsus length, wing length, and ninth primary feather length, indicated that development had not been adversely affected. Regression analyses of rate of weight gain were not significantly different between exposed and control nestlings. Deposit of insecticide at ground level, as measured by glass plate rinsings, was extremely low and variable across the spray block.

The findings suggest that Zectran® , when applied at operational dosage rates, should not adversely affect White-throated Sparrow nestlings. However, observations of apparently lethargic and unresponsive nestlings soon after the spray raise questions about the validity of using nestling growth and development to assess impact of carbamate insecticides on songbirds. Further, it is recommended that a double dose spray be studied to gauge the margin of safety of Zectran® to nestling White-throated Sparrows, a species amenable to study and considered to be a good indicator of songbirds in general.

Résumé

On soumet actuellement à des tests l'insecticide Zectran® , un inhibiteur de la cholinestérase, qui est employé au Canada dans la lutte contre la tordeuse du bourgeon de l'épinette (Choristoneura fumiferana). A cause de la toxicité relativement élevée du produit et des dangers que présente son ingestion pour les oiseaux, le Service canadien de la faune (région de l'Atlantique), alarmé par l'enregistrement du Zectran®, a tenté de déterminer si l'arrosage au Zectran® présente un danger pour les oiseaux chanteurs. Le Service canadien de la faune s'inquiète particulièrement des oisillons qui sont plus sensibles aux insecticides inhibiteurs de la cholinestérase que ne le sont les oiseaux chanteurs adultes.

On a donc suivi la croissance et le développement des oisillons de Pinsons à gorge blanche dans un secteur ayant fait l'objet d'un arrosage aérien au Zectran® (un insecticide du groupe des carbamates, matière active, mexacarbate). On a comparé les moyennes établies chaque jour selon quatre paramètres soit le poids, la longueur du torse, la longueur de l'aile et la longueur de la neuvième rémige primaire et ce pour les oisillons ayant été exposés aux insecticides comme pour le groupe contrôle. Il ressort de la comparaison des divers paramètres que le développement des oisillons n'a pas été affecté par les insecticides. Pour ce qui est du taux d'augmentation de poids, les analyses de la régression ne diffèrent pas considérablement d'un groupe à l'autre. On a découvert très peu de dépôts d'insecticides au sol, à partir de l'analyse de la solution de rinçage des plaques de verre, et les dépôts variaient d'ailleurs beaucoup sur toute la surface arrosée.

Les résultats de l'étude suggèrent que lorsque le Zectran® est appliqué selon le dosage opérationnel, il ne devrait pas nuire aux oisillons de Pinsons à gorge blanche. Cependant après avoir aperçu, peu après l'arrosage, des oisillons qui semblaient léthargiques et qui réagissaient peu au stimulus, on a tendance à remettre en cause la validité des études qui se fondent sur la croissance et le développement des oisillons pour évaluer l'incidence des insecticides du groupe des carbamates sur les oiseaux chanteurs. Le rapport recommande en outre que l'on étudie les effets de l'arrosage d'une double dose de Zectran® afin d'établir la marge de sécurité du produit pour les oisillons des Pinsons à gorge blanche qui se prêtent bien à l'étude et qui sont considérés comme de bons indicateurs de l'état des oiseaux chanteurs en général.

Since 1952 the province of New Brunswick has depended upon the use of chemical insecticides to control spruce budworm (Choristoneura fumiferana) damage to the coniferous forest. Chemicals in current use are cholinesterase (ChE) inhibitors which act by disrupting nervous system functions of the budworm and result in loss of respiratory ability and eventual death. Vertebrates, too, are susceptible to exposure to ChE-inhibiting chemicals through disruption of their nervous systems. Especially vulnerable are the canopy-dwelling songbirds which use the same habitat as the target organism and whose fast-paced life relies heavily upon an efficiently-functioning nervous system.

The ChE-inhibiting insecticide Zectran® UCZF 19 (active ingredient mexacarbate) is currently being tested for spruce budworm control in Canada and has potential for widespread use. Because of its relatively high acute oral toxicity to birds (Hudson et al. 1984), concern about its possible use prompted the Canadian Wildlife Service (Atlantic Region) to determine if field application was hazardous to songbirds. Of primary concern is the health of nestlings: operational spraying against budworm takes place at the time when many songbirds are rearing young and, because ChE activity is very low and develops only slowly during the nestling period (Grue et al. 1981; Grue and Hunter 1984), nestlings would be least able to tolerate ChE inhibition. Also, they would be the most affected by disruptions in the available food supply.

The present study was conducted to determine if Zectran® UCZF 19 affected the growth and development of White-throated Sparrow (Zonotrichia albicollis) nestlings when applied aerially at a dosage rate of 70 g AI (active ingredient)/ha, the probable operational dose if use of the product is authorized.

Study Area

The study area was located within the Harcourt District of the Maritime Lowlands Ecoregion (Loucks 1962), approximately 17 km W of Harcourt, New Brunswick. Gently rolling hills support a coniferous forest composed primarily of balsam fir (Abies balsamea), black spruce (Picea mariana), red spruce (P. rubens), and eastern white pine (Pinus strobus). Some deciduous growth, including red maple (Acer rubrum), white birch (Betula papyrifera), and yellow birch (B. alleghaniensis), is dispersed throughout. The sprayed site was rectangular and 0.65 km X 1.2 km in size. The boundaries of the untreated control site were not defined but no nests were nearer than 2 km to the treated area.

Both study sites had been clear cut approximately 10 years earlier. Regeneration consisted essentially of balsam fir, white birch, trembling aspen (Populus tremuloides), and cherry (Prunus spp.). Ground cover was dominated by bracken fern (Pteridium aquilinum), haircap moss (Polytrichum commune), sphagnum moss (Sphagnum spp.), raspberry (Rubus sp.), and rhodora (Rhododendron canadense).

Residual trembling aspen, white birch and red maple were scattered throughout the study sites.

Methods

The White-throated Sparrow was chosen as a representative and suitable songbird for detailed study. Its breeding biology has been thoroughly researched and is relatively well understood; it occurs widely and abundantly throughout New Brunswick, allowing researchers great flexibility in responding to experimental insecticide trials, the location of which is often unknown until after planning has begun. The White-throated Sparrow nests on the ground and is relatively tolerant of human activity, allowing frequent nest checks with a low probability of desertion. In New Brunswick, its breeding densities are greatest in clear-cut areas where exposure to sprays might be similar to that of canopy-dwelling birds which tend to be most affected by forest spraying (Moulding 1976; Zinkl et al. 1977; Pearce et al. 1979) but which are less amenable to study.

Fieldwork began on 27 May 1985. Nests were found by intensively searching both the experimental and control sites. Once found, the nests were checked regularly, usually once a day in the morning. After the eggs hatched, the following growth measurements were taken daily: 1) weight, 2) tarsus length, 3) wing length, and 4) ninth primary length. Weight was taken with a 30 g Pesola scale, and estimated to the nearest 0.1 g. The other measurements were taken with a Vernier caliper to the nearest 0.05 mm. All measurements were taken between 0830 hr and 1300 hr. An effort was made to visit a given nest at the same time each day to reduce the variability that might be attributed to the timing of the visit. The timing of nests visits did not vary by more than 2 hours, and usually less than 1 hour.

Measurements were continued until the nestlings fledged, usually Day 8 or 9, with day of hatch being considered Day 0. Fieldwork ended on 30 June 1985. It was planned that the experimental site would receive two treatments of Zectran®, five days apart, timed to expose the maximum number of young nestlings to both sprays, a worst-case scenario. Once the eggs began to hatch the calculated best date for the first spray was determined to be 14 June. Because of poor weather and mechanical problems the first spray was delayed by 2 days. The first treatment of Zectran® was given on the morning of 16 June starting at 0620 hr. The second spray was, as planned, 5 days after the first, on the morning of 21 June beginning at 0635 hr. Spray operations took 20 to 30 minutes to complete. Both treatments were made by Cessna 188 Ag truck aircraft equipped with 4 Micronair® AU3000 rotary atomizers. The spray was emitted at a dosage rate of 70 g AI/ha; the formulation consisted, by volume, of 22% Zectran®, 3% Triton® X 114 (emulsifier) and 75% water (carrier), and was sprayed at rate of 1.5 L/ha.

An attempt was made to measure spray deposit at ground level. Glass plates, each 0.1 m^2, were placed along a logging road crossing the study area, at about 30 m intervals. One hour after spraying each plate was thoroughly rinsed with ethyl acetate into an amber-coloured jar. Analysis of the rinsings for Zectran® was performed at the laboratories of the New Brunswick Research and Productivity Council at Fredericton, by gas/liquid chromatography.

Significance for all statistical tests was set at the 0.05 level. Procedures for regression analyses are from Zar (1974).

Results

Ten experimental and 6 control nests provided data relevant to this study. Of the experimental nests, 9 had nestlings aged between Day 4 and 7 on the day of the first spray. Although there is usually a 9-day nestling period, the 5-day interval between sprays resulted in only 1 nest, with 2 nestlings (both Day 9), being exposed to the second spray. The one nest which had eggs on 16 June was considered to have not been exposed to the first spray. It contained Day 1 nestlings during the second spray. Because only 5 experimental and 2 control nestlings remained in the nest till Day 9, that day was dropped from the statistical analyses. All the experimental nestlings in this study can therefore be considered to have received a single exposure to Zectran® spray delivered at a dose of 70 g AI/ha. The number of experimental nestlings exposed at each age is given in Table 1.

The 30 nestlings exposed to the Zectran® do not include 2 which are part of a nest of 3 young on the day prior to the spray, but which were missing when that nest was checked about 2 hours after the spray. The single remaining nestling in that nest as well as 2 nestlings in another nest looked lethargic and sickly on the morning of the spray. Those three nestlings seemed not to struggle or gape as much as other nestlings of that age would normally do. They appeared to have recovered by the following day. No other exposed nestlings exhibited those changes in behaviour during any post-spray visit to a nest. No control nestlings showed any of the behaviours described above.

The effect of the Zectran® spray on the growth of nestling White-throated Sparrows was examined in two ways. First, the daily means of the four body parameters measured in experimental nestlings were compared with controls (Table 2). Of the 34 comparisons of the means by unpaired t-tests, only one, tarsus length on Day 1, showed a significant difference. The daily change in the means of each body parameter measured, for experimentals and controls, is graphically presented in Figure 1 (a-d). The second method involved a more detailed analysis of the rate of weight gain. Because of asynchronous hatching, the nestlings were not all the same age when they were exposed to the pesticide. In previous work it was found that tarsus development was least likely to be affected by sub-lethal exposure to pesticides. In another study it was determined that when hatching time

Table 1. Age distribution of nestling White-throated Sparrows subjected to a single exposure of Zectran® sprayed at a dosage rate of 70 g AI/ha.

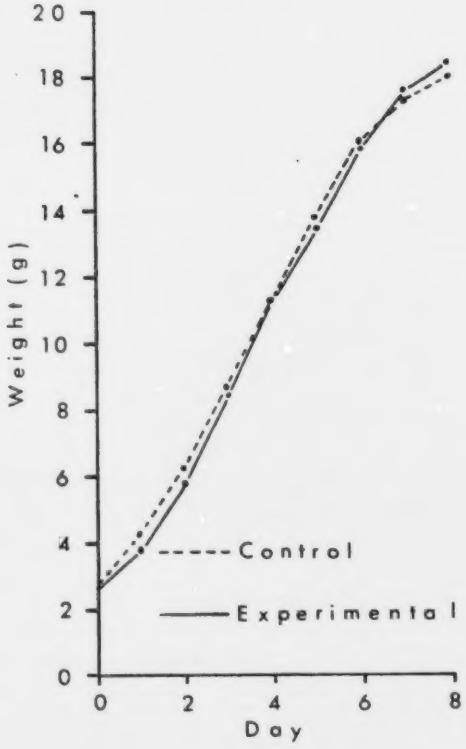
Age (Day)	Number of exposed nestlings
1	5
4	10
5	4
6	4
7	7
Total	30

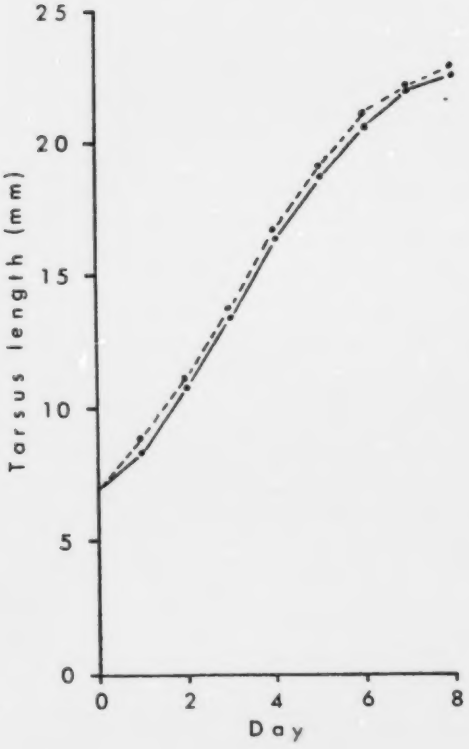
Table 2. Summary of daily growth measurements (mean, SD, n) of exposed (E) and control (C) nestling White-throated Sparrows.

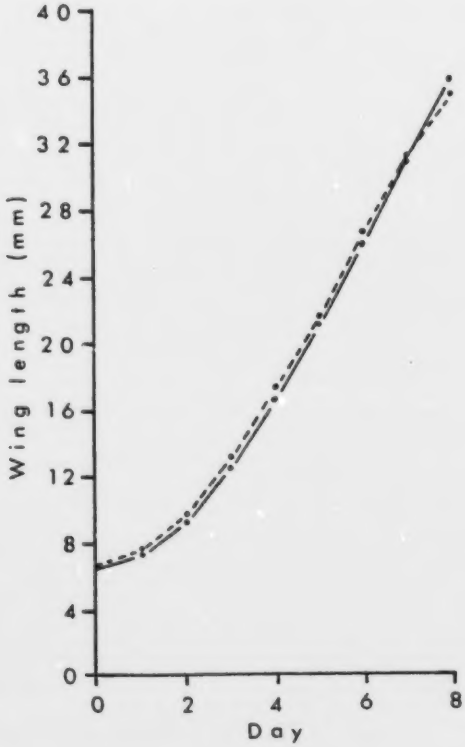
nestling age	weight (g)		tarsus (mm)		wing (mm)		9th primary (mm)	
	E	C	E	C	E	C	E	C
Day 0	2.6	2.7	7.0	7.0	6.6	6.6	-	-
	0.3	0.4	0.4	0.5	0.4	0.4	-	-
	20	20	20	20	20	20	-	-
Day 1	3.9	4.2	8.3*	8.8*	7.5	7.8	-	-
	0.5	0.6	0.6	0.6	0.4	0.5	-	-
	20	21	20	21	20	21	-	-
Day 2	5.8	6.2	10.8	11.1	9.4	9.7	0.5	0.5
	0.6	0.9	0.8	0.9	0.8	0.9	0.1	0.2
	32	23	32	23	32	23	7	4
Day 3	8.5	8.7	13.4	13.7	12.5	12.9	1.0	1.2
	0.9	1.2	1.0	1.0	1.1	1.4	0.4	0.4
	31	23	31	23	31	23	30	23
Day 4	11.3	11.3	16.3	16.5	16.5	16.8	2.8	3.1
	1.4	1.4	1.1	1.2	1.4	1.5	0.8	0.8
	30	23	30	23	29	23	30	23
Day 5	13.5	13.7	18.6	19.1	21.1	21.5	5.8	6.3
	1.2	1.4	1.1	1.1	1.6	2.0	1.1	1.3
	30	22	30	22	30	22	30	22
Day 6	15.9	16.0	20.6	21.0	26.1	26.6	9.6	10.0
	1.3	1.1	0.9	0.8	1.9	2.1	1.2	1.4
	30	19	30	19	30	19	30	19
Day 7	17.6	17.4	21.8	21.9	31.3	30.8	13.2	12.8
	1.0	1.2	0.7	0.7	2.0	2.3	1.1	1.6
	30	13	30	13	30	13	30	13
Day 8	18.3	17.9	22.4	22.7	35.8	34.6	16.5	16.0
	1.0	1.3	0.6	0.5	2.2	2.3	1.2	1.8
	25	7	25	7	25	7	25	7

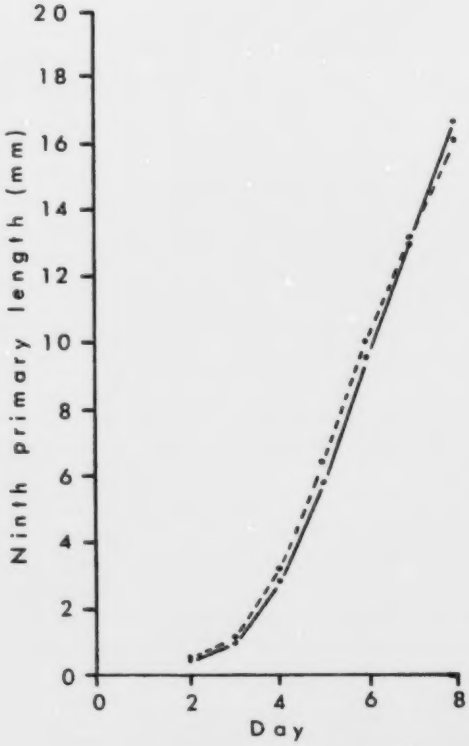
*indicates significant difference between exposed and control sample means

Figure 1. Growth of White-throated Sparrow nestlings in experimental (sprayed) and control (unsprayed) areas. (a) weight, (b) tarsus length, (c) wing length, (d) ninth primary length.


Weight (g)
Day
Control
Experimental


Tarsus length (mm)
Day


Wing length (mm)
Day


Ninth primary length (mm)
Day

was accurately known, tarsus length was the body parameter most closely correlated to age (unpublished data). (In a stepwise multiple regression with weight, tarsus length, wing length, and outer primary length as independent variables, tarsus length was picked in the first step and accounted for about 95% of the variance in age.) Therefore, tarsus length, rather than age, was used as the independent variable in the following analysis of the rate of weight gain. From a visual inspection of the growth curve it was determined that tarsus growth was nearly linear between Days 2 and 7 inclusive. A linear regression program was used to calculate the slope (= rate) of the weight gain curve.

Five regression equations were calculated for nestlings aged from Day 2 to 7. They represent: 1) all control nestlings, 2) pre-spray experimentals, 3) experimentals, day of spray, 4) experimentals, 1 day after spray, and 5) experimentals, 2 days after spray. A multiple comparison of the slopes of the regression lines showed no significant difference in the rates of weight gain among controls, pre-spray experimentals, and post-spray experimentals (Table 3).

The result of deposit measurement are presented in Table 4. An average of 1.9% of the active ingredient emitted per unit area was recovered on the plates after the first spray and 4% was recovered after the second application. There was a high degree of variability from plate to plate on both sprays.

Discussion

During the past decade the use of relatively non-persistent insecticides has increased markedly, primarily due to their low potential to accumulate in biological systems (Lamoreaux and Newland 1977). The high acute toxicity of those chemicals, however, may result in mortality to wildlife immediately following or relatively soon after application (see Grue *et al*. 1983; Hill and Fleming 1982). Sub-lethal impacts also have been described, usually relating to pesticide-induced anorexia (Grue 1982). Sub-lethal impacts of organo-phosphorus insecticides on wildlife have been relatively thoroughly studied (e.g. Powell 1984; Grue and Shipley 1984; Grue *et al*. 1982). The effects of carbamate insecticides on wildlife are, however, less well studied and understood. They, too, may be highly acutely toxic, and potential for non-target impact is considerable.

The present study indicates that application of Zectran® for spruce budworm control should have no serious effects on nestling White-throated Sparrows when applied at recommended rates. None of the growth parameters measured was affected and no mortality was believed to be a consequence of exposure to the insecticide. Fledging weights, thought to be particularly important in post-fledging survival (Perrins 1965; Smith 1967), showed no significant difference when the exposed birds were compared to controls. Other body parts, such as the tarsus, wing, and outer primary, were also of normal size at fledging. The

Table 3. Summary of multiple regression comparisons of the slopes (rates) of weight gain of White-throated Sparrows as a function of tarsus length.

	intercept	slope	residual ss	residual df
Control	-4.8412	0.9892	76.919	121
Experimental:				
pre-spray	-5.1436	1.0155	30.245	84
day of spray	-4.5077	0.9978	4.440	16
1 day post-spray	-5.7336	1.0309	8.015	16
2 days post-spray	-7.8111	1.1584	4.156	12

F-value = 0.4501, $p > 0.05$, df = 4 and 249.

Table 4. Deposition of mexacarbate (g/ha) on glass plates in study area sprayed at a dosage rate of 70 g/ha by aerial application.

Plate #	Spray 1	Spray 2
1	4.4	3.3
2	1.6	2.3
3	0.6	2.3
4	0.4	3.6
5	1.0	5.3
6	0.8	5.3
7	0.5	3.0
8	0.7	2.1
9	1.1	0.4
10	2.0	0.2
Average	1.3	2.8

single significant difference in tarsus means on Day 1 can be explained as a random occurrence. In 34 comparisons at the 0.05 probability level, 1 to 2 tests can be expected to be significant by chance alone.

One incident which cannot be discounted as an insecticide-induced impact involved 3 nestlings, from 2 nests, that did not appear as healthy and active as other nestlings of similar age in the control area on the morning of the first spray. There was also a noticeable absence of gaping, a behaviour normally exhibited by nestlings when handled. Although those 3 nestlings appeared to have recovered by the next day, manifestation of behavioural abnormalities symptomatic of poisoning (Grue and Shipley 1984) suggests the margin of safety of Zectran® application to nestlings is narrow.

Even though some impact may have occurred as a result of the spray, growth of nestlings was apparently not affected. Similar results were also noted in a study of the effects of orally-administered aminocarb, another carbamate insecticide, on White-throated Sparrow nestlings (unpublished data); unless the nestling died, there was no measurable impact on growth. That is in marked contrast to the effects of an organophosphate insecticide, fenitrothion, on nestlings, in which growth was measurably affected at sub-lethal doses (unpublished data). Although both insecticides are ChE inhibitors (O'Brien 1967), differences in dose-response may be related to the mode of recovery from exposure. ChE inhibition induced by carbamates is a readily and rapidly reversible chemical reaction (Murphy 1975). Recovery thus occurs by reversibility of the initial reaction as well as by de novo synthesis of ChE. Recovery from organophosphate poisoning usually occurs only from the latter process (Fleming 1981). Thus, unless exposure is initially sufficient to induce death, recovery from exposure to carbamates may be so rapid as not to be measurable in terms of growth impairment.

Deposit of insecticides on the glass plates was extremely light in spite of the near-ideal weather conditions at the time of both applications. Rinsing of plates 1 hour after spraying should have prevented significant photo-degradation of the parent compound, especially as the plates were not subjected to direct sunlight at that early hour. However, there may be two problems with the assessment technique: 1) 1 hour may be insufficient time for complete deposition to occur, and 2) a flat glass surface may not be a suitable receptor for fine spray droplets. Further evaluation of deposit assessment techniques is required so that bio-effects researchers can better relate their findings to the real-life spray application, as distinct from the theoretical ideal.

Although apparently relatively safe when applied at operational dosage rates, some concern about the risk of Zectran® to nestling songbirds must be expressed. Due to imprecision in aircraft guidance, operational spraying may result in some overswathing, a phenomenon known to have serious consequences to songbirds (Busby et al. 1983) exposed to fenitrothion. The magnitude and complexity of the

New Brunswick forest spray program render quantification of the frequency of overswathing virtually impossible. To ensure the health of songbird nestlings it is therefore advisable that any insecticide be tested and determined to be safe at double the operational dosage rate, as Zectran® was with regard to adult songbirds (in prep.). Information on the magnitude and persistence of mexacarbate residues in the food supply of songbirds would also help to define the risk of Zectran® spraying to adult and nestling birds.

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